

Effect of restricted forage intake on ruminal disappearance of bromegrass hay and a blood meal, feather meal, and fish meal supplement¹

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ABSTRACT: Two experiments were conducted to determine in situ disappearance of bromegrass hay and a ruminally undegraded protein (RUP) supplement in beef cattle fed restricted amounts of forage. Six Angus crossbred cattle (BW = 589 ± 44.4 kg; three steers and three heifers) fitted with ruminal cannulas were fed chopped (2.54 cm) bromegrass hay (8.9% CP) at one of three percentages of maintenance intake (30, 55, or 80%; one steer and one heifer per treatment). In both experiments, the cattle were allowed 7 d for diet adaptation followed by 3 d of sample collection. In Exp 1, in situ bags (50 µm pore size) containing 4.1 g of bromegrass hay (OM basis) were inserted into the rumen and subsequently removed at 3, 6, 9, 12, 15, 18, 24, 36, and 48 h after insertion. Nonlinear regression models were used to determine the rapidly solubilized protein Fraction A, the potentially ruminal degradable protein Fraction B, the ruminally undegraded protein Fraction C, and protein degradation rate. Intake level did not affect ($P = 0.15$ to 0.95) forage protein remaining after in situ incubation or Fractions A, B, and C; however, effective ruminal degradation of hay protein tended to increase quadratically ($P = 0.12$) as forage intake increased. In Exp 2, 4.2 g (OM basis) of an RUP supplement (6.8%

porcine blood meal, 24.5% hydrolyzed feather meal, and 68.7% menhaden fish meal) formulated to provide equal amounts of metabolizable protein across all levels of hay consumption was evaluated in a similar manner as in Exp 1. The undegraded protein fraction of the supplement did not differ ($P = 0.16$ to 0.74) across treatments at 3, 6, 9, and 18 h; however, increasing forage intake resulted in a linear increase ($P \leq 0.06$) in undegraded protein remaining at 12, 15, 24, 36, and 48 h. Dietary treatment had no effect ($P = 0.30$) on protein Fractions A, B, or C; however, protein degradation rate of the supplement decreased linearly ($P = 0.03$) as forage intake increased. Therefore, effective ruminal degradation of the supplement decreased linearly ($P = 0.01$) from 50.8 to 40.9% as forage intake increased from 30 to 80% of maintenance. Corresponding estimates of supplement RUP were 49.2, 56.5, and 59.1% for the 30, 55, and 80% of maintenance intake treatments, respectively. Restricting dietary intake can decrease the quantity of dietary protein that escapes ruminal degradation. Tabular estimates of RUP may not be appropriate for formulating diets to balance metabolizable protein in beef cattle consuming limited quantities of forage.

Key Words: Cattle, Effective Ruminal Degradation, In Situ, Intake, Ruminally Undegradable Protein

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Introduction

Proper amounts of protein and energy are essential for reproduction in ruminant livestock (Dunn and Moss, 1992). Most research investigating nutritional effects on reproduction has imposed restricted OM intake in an effort to decrease dietary energy (Shillo, 1992).

Scholljegerdes et al. (2004) demonstrated that essential AA flow to the duodenum decreased linearly in beef cattle fed restricted amounts of forage. Thus, nutritional effects on reproduction cannot be attributed solely to either decreased protein or insufficient energy when researchers used restricted OM intake as a method to limit dietary energy. As an initial step toward separating physiological effects of protein from energy in ruminants, metabolizable protein would need to be balanced in animals consuming restricted quantities of OM. Unfortunately, tabular values used in balancing beef cattle diets for metabolizable protein often originate from cattle that are fed for ad libitum consumption. Using these values to balance metabolizable protein supply for cattle fed severely restricted (below maintenance requirements) quantities of forage may not be

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Table 1. Ingredient and chemical composition of diets fed to cattle (Exp. 2)

Item	Treatments ^a		
	30	55	80
Ingredients, % of OM			
Bromegrass hay	52.5	77.6	90.8
Blood meal	3.2	1.5	0.6
Feather meal	11.6	5.5	2.3
Fish meal	32.7	15.4	6.3
Nutrient composition			
DM, %	93.4	93.6	93.7
OM, % of DM	87.7	89.2	90.0
% of OM			
NDF	44.6	55.0	60.3
N	7.6	4.5	2.9

^aCattle were fed 30, 55, or 80% of the forage intake required for maintenance (NRC, 2000).

appropriate because intake restriction can alter the extent of ruminal protein digestion (Scholljegerdes et al., 2004). Limited data are available on the evaluation of ruminally undegraded protein (**RUP**) of dietary ingredients for cattle fed restricted amounts of forage and high-RUP supplements. The objectives of this study were to evaluate the influence of restricted forage intake on ruminal degradability of forage and a high RUP supplement.

Materials and Methods

Six Angus-cross cattle (589 ± 44.4 kg) were fed chopped (2.54-cm) bromegrass hay (8.9% CP) during two in situ experiments. In Exp. 1, one steer and one heifer were assigned randomly to one of three forage intakes (30, 55, or 80% of maintenance) without supplement. In Exp. 2, one steer and one heifer were fed bromegrass hay at one of the three aforementioned intake levels and an RUP supplement (6.8% porcine blood meal, 24.5% hydrolyzed feather meal, and 68.7% menhaden fish meal) formulated to balance metabolizable protein across all levels of hay consumption (Table 1). The RUP supplement was offered at a level (2,556, 1,491, and 762 g/d for the 30, 55, and 80% treatments, respectively) that would supply the duodenum with approximately the same quantity of total essential AA as that of an animal consuming 105% of the forage required for maintenance. The quantity of supplement offered was determined using the equation described by Scholljegerdes et al. (2004), where total duodenal essential AA flow = $(0.0155 \times \text{OM intake}) + 1.546$. Forage intake (on an OM basis) was 0.97, 1.23, and 1.56% of BW for the 30, 55, and 80% treatments, respectively. These in situ trials were conducted to provide preliminary information that would allow for an accurate assessment of how restricted intake would influence the RUP value of a supplement. Furthermore, in situ experiments were conducted separately to avoid the influences of additional ruminally degradable protein associ-

ated with the supplement on hay protein degradation (Mathis et al., 2000). In both experiments, the cattle were allowed 7 d for diet adaptation followed by 3 d of sample collection. On d 8 in both experiments, duplicate in situ bags (Ankom Co., Fairport, NY; 50 μm pore size) containing 4.1 g of OM (ground to pass a 2-mm screen) of either bromegrass hay (Exp. 1) or 4.2 g of supplement OM (Exp. 2) were soaked in tepid water for 1 min and inserted into the ventral rumen before feeding. A single insertion time seemed ideal because samples were exposed to common ruminal conditions over the course of the incubation (Michalet-Doreau and Ould-Bah, 1992) and cattle were fed fewer than four meals per day (Vanzant et al., 1998). Total weight of supplement and bromegrass hay in the in situ samples was subtracted from the quantity of the respective ingredient for the d-8 feeding. In situ bags were removed at 3, 6, 9, 12, 15, 18, 24, 36, and 48 h after insertion into the rumen and immediately frozen at -20°C for no less than 48 h. For both experiments, in situ bags were rinsed in tepid water after the last bag was removed from the rumen and dried for 7 d in a 55°C forced-air oven. In Exp. 1, in situ residues were dried, weighed, and rinsed with NDF solution to correct for possible microbial contamination (Mass et al., 1999) and subsequently analyzed for N (Leco model FP-528 nitrogen determinator; Leco Corp., St. Joseph, MI). In situ residues from Exp. 2 were handled in the same manner as for Exp. 1, with the exception of the NDF rinse, which was not conducted due to the theoretically low microbial attachment to the supplement (Nocek, 1985).

Using the NLIN procedure of SAS (SAS Inst., Inc., Cary, NC), the model of Ørskov and McDonald (1970) was used to calculate protein Fractions A and B, as well as protein degradation rate ($k_d = \%/h$). Effective ruminal degradation (**ERD**) was calculated using the equations of Broderick (1994). Particulate passage rate (k_p) estimates of 4.62, 6.65, and 3.89%/h for the 30, 55, and 80% treatments, respectively, were obtained from Scholljegerdes et al. (2001), who fed forage levels similar to those reported herein. These particulate passage rate values were used for the calculation of forage ERD because forage would most likely be associated with the particulate phase (Coblentz et al., 1999). Fluid passage rate values of 4.44, 6.43, and 7.85%/h for 30, 55, and 80% treatments, respectively, also were obtained from the companion study, Scholljegerdes et al. (2001). Fluid passage rate estimates were used to calculate supplement ERD because the supplement would most likely be associated with the fluid phase (Nocek, 1985).

All data were analyzed by using the GLM procedures of SAS as a completely randomized design one-way ANOVA. Gender was not included in the statistical model. Single degree of freedom polynomial contrasts were used to determine linear and quadratic effects of level of feed intake (Steel and Torrie, 1980).

Results and Discussion

Forage residual protein was not affected ($P = 0.15$ to 0.93) by level of forage intake at any incubation time

Table 2. Effect of restricted intake on protein degradability characteristics of brome-grass hay

Time/item	Treatments ^a			SEM ^b	Contrasts	
	30	55	80		Linear	Quadratic
	— % protein remaining —					
3	68.1	65.9	63.0	2.2	0.15	0.93
6	61.3	59.7	60.6	2.9	0.87	0.73
9	56.1	52.8	51.5	2.1	0.17	0.71
12	46.7	42.2	45.2	4.0	0.80	0.48
15	40.0	37.9	37.6	1.9	0.39	0.71
18	36.9	33.8	33.7	5.2	0.38	0.67
24	30.5	32.3	28.6	3.6	0.57	0.40
36	25.1	23.2	25.8	2.1	0.83	0.41
48	20.9	21.8	21.4	2.0	0.87	0.80
Fraction A, % OM	19.3	20.4	23.8	3.6	0.40	0.80
Fraction B, % OM	62.8	60.8	58.2	4.4	0.39	0.95
Fraction C, % OM	17.9	18.8	18.0	0.8	0.92	0.43
k _d , %/h	6.5	7.4	6.6	0.4	0.78	0.17
ERD ^c	55.9	52.3	60.3	1.7	0.12	0.03
RUP ^d	44.1	47.7	39.7	1.7	0.12	0.03

^aCattle were fed either 30, 55, or 80% of the forage intake required for maintenance (NRC, 2000).

^bn = 2.

^cEffective ruminal degradation = % Fraction A + [% Fraction B × (k_d/(k_d+k_p))] where k_p = 4.44, 6.65, and 8.89%/h for the 30, 55, and 80% treatments, respectively.

^dRuminally undegradable protein = 100 – ERD.

(Table 2). These data are comparable to our in vivo data, in which Scholljegerdes et al. (2004) reported that ruminal protein degradability was not affected by feeding beef heifers forage at levels similar to those reported herein. Forage protein Fractions A, B, and C did not differ ($P = 0.39$ to 0.95) across levels of forage intake, suggesting that intake did not influence the ruminal microbes and the extent to which they degrade these fractions. Forage protein k_d was not influenced ($P = 0.17$) by forage intake and averaged 6.8%/h across intake levels. Our estimates of forage k_d are within the range of 5 to 8%/h reported by other researchers (Broderick, 1994; Varel and Kreikemeier, 1995; Coblenz et al., 1999). Although the rate of degradation was not affected by restricted intake, the ERD of forage N increased ($P = 0.03$) quadratically, with the ERD increasing from 30 to 55%, and then decreasing for the 80% treatment. There was a concomitant quadratic decrease ($P = 0.03$) in the RUP fraction with an increase in forage intake from 30 to 80% of maintenance. The quadratic increase in ERD is likely an artifact of the cubic trend in passage rate (k_p = %/h) reported by Scholljegerdes et al. (2001). Calculating forage ERD based on estimated k_p from the equations reported in the NRC (2000; Table 3) resulted in a linear decrease ($P = 0.03$) in ERD. Additionally, if one calculates the ruminal residence time of the protein by taking the inverse of k_p (1/k_p; Ellis, 1978) and applies the actual k_p values reported by Scholljegerdes et al. (2001), the calculated time points are 21.6, 15.0, and 25.7 h for 30, 55 and 80%, respectively. The protein remaining at these approximate times (Table 2) indicates that both the actual and NRC closely represent the extent of protein degradabil-

ity at their respective time points. Therefore, the use of NRC calculated k_p would be appropriate when actual k_p values are not available for evaluation of how intake might influence ruminal degradation rates of forage protein.

In Exp. 2, restricting forage intake did not influence ($P = 0.16$ to 0.74) supplement protein remaining at 3, 6, 9, and 18 h (Table 4); however, supplemental protein remaining at 12, 15, 24, 36, and 48 h increased linearly ($P \leq 0.06$) as intake increased from 30 to 80% of maintenance. Greater supplemental protein degradation in cattle fed restricted amounts of forage is likely associated with increased ruminal retention time, which would allow the ruminal microbes more time to degrade the protein (Shadt et al., 1999). Supplement k_d decreased ($P = 0.03$) linearly as forage intake increased. Data for the k_d of our ingredients is unavailable in restricted-fed animals, as well as animals fed an all-forage diet. Nevertheless, based on published values for the k_d of blood meal (0.0%/h; Maiga et al., 1996), feather meal (0.4%/h; Broderick et al., 1988), and fish meal (1.4%/h; Broderick et al., 1988), we would calculate our k_d of the RUP supplement to be 1.06%/h. A k_d of 1.06%/h is similar to what we determined in the current trial, which demonstrates that our values are not unreasonable for these particular ingredients. The ERD of our supplement decreased linearly ($P = 0.01$) and RUP increased ($P = 0.01$) concomitantly as intake went from 30 to 80% of maintenance. The decreased ERD for the supplement is likely related to increased ruminal retention with increased severity with forage intake restriction, with a resultant increase in ruminal degradation of the RUP supplement. The increase in

Table 3. Effect of restricted intake on the effective ruminal degradation of bromegrass hay calculated using either actual or NRC (2000) calculated k_p

Item	Treatments ^a			SEM ^b	Contrasts	
	30	55	80		Linear	Quadratic
Actual						
k_p^c	4.6	6.7	3.9	0.02	0.001	0.001
ERD ^d	50.9	52.3	60.3	1.7	0.12	0.03
NRC ^e						
k_p^c	1.9	2.8	3.5	0.02	0.001	0.01
ERD ^d	67.7	64.3	61.7	1.5	0.03	0.82

^aTreatments = % of the forage intake required for maintenance according to the NRC (2000).

^bn = 2.

^c k_p = %/h.

^dEffective ruminal degradation = % Fraction A + [% Fraction B \times [$k_d/(k_d+k_p)$]].

^eCalculated k_p based on the equation of the NRC, 2000: $(0.388 + [0.022 \times \text{DMI/SBW}^{0.75}]) + 2.0 \times \text{Forage DM in DMI, \%}^2/100$.

ruminal degradation at the more severely restricted intake level is contrary to what was noted for the forage. These differences may be due to the lack of nutrients available to the cellulolytic bacteria when intake levels are severely restricted (Scholljegerdes et al., 2004). Nonetheless, the increase in ruminal degradation of the RUP supplement could be due to the combination of a decrease in passage rate with the additional ruminally degraded protein being supplied by the RUP supplement affording the microbes more nutrients, thereby allowing for greater ruminal degradation of protein that is often seen when intake is limited (Merchen et al., 1986).

Tabular values for RUP found in the NRC (2000) suggest that the RUP of this supplement would be

64.1%; however, when intake was restricted from 80 to 30%, the actual RUP values for the supplement ranged from 59.1 to 49.2%, which are well below the NRC-tabulated value. Based on the averages for the forage and RUP supplement, the overall dietary RUP (forage plus RUP supplement) was 42.8, 45.9, and 42.8% for the 30, 55, and 80% treatments, respectively. The evaluation of the dietary RUP value using the NRC (2000) Level II computer model would indicate that the dietary RUP of 58.3, 57.4, and 51.8% for the 30, 55, and 80% of maintenance, respectively. The decrease in dietary RUP value based on NRC (2000) calculations is plausible because the quantity of RUP supplement decreased as forage intake increased. However, in comparison with the actual RUP calculations, it would seem that

Table 4. Effects of restricted intake on in situ protein degradability characteristics of a combination of blood meal, feather meal, and fish meal

Time/item	Treatments ^a			SEM ^b	Contrasts	
	30	55	80		Linear	Quadratic
	% protein remaining					
3	61.7	64.2	63.5	0.7	0.16	0.16
6	60.3	60.4	62.0	1.5	0.49	0.70
9	58.0	59.2	59.4	1.2	0.47	0.74
12	56.3	58.6	59.1	0.6	0.05	0.35
15	54.5	59.3	58.6	0.6	0.02	0.04
18	51.4	55.0	56.2	2.5	0.27	0.72
24	44.9	51.7	55.0	1.8	0.03	0.48
36	36.7	42.0	48.6	2.4	0.04	0.86
48	31.5	35.7	42.5	2.6	0.06	0.71
Fraction A, %	33.5	32.6	34.6	0.9	0.45	0.30
Fraction B, %	66.5	67.4	65.4	0.9	0.45	0.30
Fraction C, %	0.0	0.0	0.0	0.0	—	—
k_d , %/h	1.6	1.2	0.8	0.1	0.03	0.81
ERD ^c	50.8	43.5	40.9	1.2	0.01	0.21
RUP ^d	49.2	56.5	59.1	1.2	0.01	0.21

^aCattle were fed either 30, 55, or 80% of the forage intake required for maintenance (NRC, 2000) plus 2,556, 1,491, or 762 g/d, respectively, of a high-RUP supplement that contained 6.8% blood meal, 24.5% feather meal, and 68.7% fish meal.

^bn = 2.

^cEffective ruminal degradation = % Fraction A + [% Fraction B \times [$k_d/(k_d+k_p)$]] where k_p = 4.44, 6.43, and 7.85%/h for the 30, 55, and 80% treatments, respectively.

^dRuminally undegradable protein = 100 – ERD.

although the NRC does account for changes in intake when calculating passage rate, it does not account for these changes when calculating the degradation constants for dietary ingredients. Therefore, the NRC (2000) model overestimates the RUP value of a diet when dietary intake is restricted. Our calculations for dietary RUP did not decrease in a linear fashion but responded quadratically. This quadratic response might indicate that no matter how long forage resides within the rumen, a portion of that protein may not be available for microbial degradation. Overall, the use of tabular values for RUP may not accurately account for the changes that occur within the rumen, especially during times of severe dietary restriction.

Recent concerns regarding feeding of mammalian protein sources to ruminants and the proposed link to the development of bovine spongiform encephalopathy may restrict the use of blood meal and similar ruminant-derived protein sources as RUP supplements for ruminants. To date, however, animal protein sources such as blood meal are not banned in the United States for use in ruminant feeds according to the FDA (Title 21 Sec. 589.2000). Nonetheless, recent deliberations by the FDA Center of Veterinary Medicine have proposed the addition of all blood-derived products, including porcine and poultry, to the prohibited feeds list. Thus, the future use of our experimental model in the study of nutrition/reproduction interactions will necessitate reformulation of the RUP supplement used in the current study. Menhaden fish meal and hydrolyzed feather meal will remain as prominent ingredients in our supplement because of their high RUP value, excellent essential AA profile, and high postruminal AA availability.

Implications

Caution must be exercised when using tabular values for ruminally undegradable protein when formulating diets for cattle consuming restricted levels of forage. An adjustment factor, including passage rate, should be used to account for increased ruminal degradation of the diet.

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